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Maintenance therapy with proton pump inhibitors and risk of gastric cancer: a nationwide population-based cohort study in Sweden

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Running title: Proton pump inhibitors and risk of gastric cancer

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Abstract (299 words)

Objective: Proton pump inhibitors (PPIs) are among the most commonly prescribed drugs. Concerns have been raised about a potentially increased risk of gastric cancer following long-term use. Our aim is to assess the risk of gastric cancer associated with PPI use, taking into account underlying indications.

Design: Population-based cohort study. Standardized incidence ratios (SIRs) and 95% confidence intervals (CIs) were calculated to compare the risk of gastric cancer among long-term PPI-users with the corresponding background population, while taking confounding by indication into account.

Setting: Population-based study in Sweden (2005-2012)

Participants: this study included virtually all adults residing in Sweden exposed to maintenance therapy with PPIs.

Exposure/Intervention: maintenance use of PPIs, defined as at least 180 days during the study period. Maintenance use of H2-receptor antagonist was evaluated for comparison reasons.

Outcome measures: gastric cancer (cardia and non-cardia), and subgroup analysis for gastric adenocarcinoma, as defined by the Swedish Cancer Register.

Results: Among 797,067 individuals on maintenance PPI therapy, the SIR of gastric cancer was over 3-fold increased (SIR=3.38, 95% CI 3.23-3.53). Increased SIRs were found in both sexes and all age groups, but were especially increased among PPI-users younger than 40 years (SIR=22.76, 95% CI 15.94-31.52). Increased SIRs were found for each indication studied,

including those without an association with gastric cancer, e.g. gastro-esophageal reflux (SIR=3.04, 95% CI 2.80-3.31), and those with a supposedly decreased risk, e.g. aspirin users (SIR=1.93, 95% CI 1.70-2.18). The association was similar for cardia and non-cardia gastric cancer. Analyses restricted to adenocarcinoma showed similar results to those for all gastric cancers. Long-term users of histamine-2 receptor antagonists, which have the same indications as PPIs, were not at any increased risk.

Conclusions: Long-term PPI-use might be an independent risk factor for gastric cancer. This challenges broad maintenance PPI therapy, particularly if the indication is weak.

Article summary Strengths and limitations of this study

- Population-based and nationwide design based on contemporary use of proton-pump inhibitors (PPIs) – resulting in sufficient power to assess underlying indications (to assess confounding by indicatin).
- To our knowledge, this is the largest study to date assessing the association between PPIs and gastric cancer
- The findings are standardized for age, sex – which are often described the major confounding factors in epidemiologic studies - and calendar time. Yet, other confounders could not be taken into account because the information was not available for the background population.
- Exposure information is based on the Swedish Prescribed Drug Registry, which is initiated in July 2005 and has a complete nationwide coverage.

Introduction

Proton pump inhibitors (PPIs), introduced in the 1980s, are potent gastric acid suppressors that reduce gastric acidity.(1-3) PPIs are among the most prescribed drugs globally,(4) used for healing peptic ulcers, counteracting gastro-esophageal reflux, eradicating *Helicobacter pylori* (in combination with antibiotics) and preventing primary or recurrent peptic ulcers, e.g. in individuals exposed to aspirin or other non-steroidal anti-inflammatory drugs (NSAIDs) or with Zollinger-Ellison syndrome (a gastrin-secreting pancreatic tumor). However, it has been suggested that long-term PPI-use increases the risk of pre-malignant gastric lesions (e.g. polyps, atrophy, and metaplasia) and gastric cancer.(2, 5, 6) Gastric acid secretion blockage may disrupt the gastric microbiome, interfere with nitrosamine formation, cause chronic atrophic gastritis and increase gastrin serum levels, which can all contribute to gastric cancer development.(2, 5, 7, 8). The effect of PPI use on the gut microbiome may even be more prominent than the effects of antibiotics.(9) Among three recent meta-analyses on the topic, one found no association between long-term PPI-use and pre-malignant gastric lesions, based on six randomized controlled trials (1789 patients in total).(2) The second included an additional trial (2343 patients in total), and found no evidence of gastric tumor development in PPI-users with atrophy or intestinal metaplasia, while an increased risk of gastric hyperplasia was indicated.(6) The third, based on 11 observational studies (94,558 participants), reported a 40% increase of gastric cancer among PPI-users.(5) However, the impact of confounding by indication remains unknown. The present study aimed to assess the risk of gastric cancer in long-term PPI-users in a population-based design, while

taking confounding by indication for such treatment into account. For comparison reasons, use of histamine 2 receptor antagonists (H2RAs), which are used for similar indications as PPIs, was also studied.

Results

Study participants

In total, 797,067 individuals on maintenance PPI therapy were included in the exposed cohort, resulting in 3,866,836 person-years of follow-up (mean 4.9 years). An additional 20,210 individuals had maintenance H2RA therapy, and 25,726 had both PPI and H2RA maintenance therapy. Figure 1 presents how the study participants were selected, and Table 1 shows their characteristics. Women constituted 58.5% of the PPI cohort, and a majority (66.1%) was younger than 70 years. Maintenance therapy with aspirin (34.8%) and NSAIDs (30.4%) were the most common indications for maintenance PPI use, followed by gastro-esophageal reflux (25.3%), gastro-duodenitis (13.2%) and peptic ulcer (10.0%). *Helicobacter pylori* infection was the indication in 7.3% of all participants, and dyspepsia in 5.5%. Barrett's esophagus and Zollinger-Ellison syndrome were rare indications (<1%) and therefore not eligible for separate analysis. A third (33.4%) of the participants had more than one of the listed indications, and was therefore included in more than one indication group. Overall mortality was slightly lower among PPI users (17.3%) compared to H2RA maintenance users (19.4%), while mortality in gastric cancer patients did not differ significantly.

Proton pump inhibitor use and overall risk of gastric cancer

Among all participants exposed to maintenance PPIs, 2,219 (0.28%) developed gastric cancer during follow-up. Of these, 1,652 (74.4%) had non-cardia gastric cancer and 1,943 (87.6%) had adenocarcinoma (Table 1). Long-term PPI-users were at more than a 3-fold increased SIR of gastric

cancer of any type (SIR=3.38, 95% CI 3.25-3.53) and gastric adenocarcinoma (SIR=3.38, 95% CI 3.23-3.53) (Table 2). After excluding early gastric cancers, the risk remained increased (SIR=1.61, 95% CI 1.51-1.71). The risk of gastric cancer was highest among individuals receiving PPI for shorter than 1 year (SIR=12.82, 95% CI 12.19-13.47), but there was also evidence for an increased risk up to 3 years of PPI use, yet reduced risks for use over 5 years (Table 3). The strength of association was similar for cardia (SIR=3.55, 95% CI 3.27-3.86) and non-cardia gastric cancer (SIR=3.33, 95% CI 3.17-3.50). The risk of gastric cancer was similar in men (SIR=3.65, 95% CI 3.45-3.85) and women (SIR=3.07, 95% CI 2.87-3.28). The SIR was higher in younger ages; PPI-users younger than 40 years had SIR of 22.76 (95% CI 15.94-31.52) while those who were at least 70 years old had SIR of 2.76 (95% CI 2.61-2.92) (Table 2). The subgroup analyses for sex and age showed similar results as the overall findings in separate analyses for gastric adenocarcinoma, as well as for cardia and non-cardia gastric cancer (Table 2).

Proton pump inhibitor use and risk of gastric cancer stratified by indication

The SIR of gastric cancer was increased in each of the 10 studied groups of indications for PPI therapy (Table 4). The highest SIRs were found among participants exposed to indications with a known association with gastric cancer, i.e. *Helicobacter pylori* infection and peptic ulcer. However, the SIRs were also increased for indications without any such association. The SIR for gastric cancer was 3.04 (95% CI 2.80-3.31) for gastro-esophageal reflux. The

SIR were increased also for indications where a decreased risk of gastric cancer was expected. The SIR was 1.93 (95% CI 1.70-2.18) among those using PPIs because of aspirin use without any other indication. The SIRs were generally similar for non-cardia gastric cancer. Regarding cardia cancer, higher SIRs than for gastric cancer were found for the indication gastro-esophageal reflux and among those exposed to aspirin or other NSAIDs, while the associations for *Helicobacter pylori* infection and peptic ulcer were less strong (Table 4).

Attributable risk

Of all 5,823 gastric cancer patients diagnosed during the study period, 38.1% occurred among maintenance PPI-users. Based on the overall SIR of 3.38, and a prevalence of PPI-use of 10.7% among adults, the attributable fraction among PPI-users (AF) was 70.4%, and 20.3% in the total population (PAF), assuming causality. Using the lowest SIR according to Table 4 (SIR=1.41), the corresponding proportions were 29.1% (AF) and 4.2% (PAF), respectively. Among individuals younger than 40, the prevalence of PPI-use was 3.3%, and 37.1% of all 97 gastric cancers occurred among PPI-users, corresponding to AF of 95.6% and PAF of 41.8%.

Use of histamine-2 receptor blockers only or proton pump inhibitors and histamine-2 receptor blockers and risk of gastric cancer

There were 12 and 62 cases of gastric cancer among maintenance H2RAs users only and both H2RAs and PPIs, respectively. The risk of gastric cancer was not increased in the H2RA only group (SIR=0.57, 95% CI 0.29-0.99), and

was moderately increased in the group exposed to both PPIs and H2RAs (SIR=2.09, 95% CI 1.61-2.69).

Discussion

This study provides some evidence of an increased risk of gastric cancer among maintenance PPI-users, including those who had PPI therapy for indications without any positive association with gastric cancer. Increased risks were found for both cardia and non-cardia gastric cancer, in both sexes and all age groups, although the risk was more pronounced in younger participants.

Strengths of the study include the nationwide and population-based design, the valid data on exposures, outcomes, and indications for PPI-use, and the large number of individuals exposed to maintenance treatment with PPIs. To our knowledge, this is the largest study to date on this topic,(10) and the cohort allowed analyses of differences in associations between underlying indications.

The main problems of this study are confounding, especially by indication, and reverse causality. Confounding by indication has been investigated by looking at the different indication groups, including groups without no increased risk of gastric cancer. Unfortunately, we could not identify any underlying indication for 25% of the PPI-users, and 39% of the H2RA-users. It is likely that clear indications were more readily recorded, while use for less rigorously diagnosed indications were more prone to be missing. This may indicate that PPI-users had more severe symptoms than H2RA-users, as also indicated by the higher proportion of PPI users presenting with multiple indications (34% vs. 19%). Yet, the lack of information on indication should not explain the associations for known indications – and those groups with no

apparent risk factors should be less likely to have severe gastrointestinal symptoms. Therefore, it was unexpected to still see such increased risks of gastric cancer in those groups. Unfortunately, we lacked information about some potential confounders, e.g. dietary factors, obesity, tobacco smoking and alcohol overconsumption since these are not collected in the nationwide Health Registries. However, the lack of association between H2RAs and gastric cancer argues against bias from confounding or selection from lifestyle factors or any other unknown factors. Since H2RAs were used for similar indications as PPIs (yet clearly became less popular), the lack of association between H2RAs and gastric cancer supports that the association between PPI-use and gastric cancer may be linked to the PPI medication *per se*. Yet, PPIs clearly became the first choice treatment for most indications for gastric acid suppression with almost 40 times more PPI maintenance users than H2RA maintenance users in Sweden. The high prevalence of PPI maintenance use among adults (10.7% in Sweden) also means that the large majority with recognized risk factors for gastric cancer will have received PPI treatment at some point. This clearly hampers assessment of PPI use as an independent risk factor and this may even be a larger problem in other study designs especially if indications of use cannot be assessed.

The problem of reverse causality, i.e. individuals taking PPIs because of symptoms arising from an undetected cancer, should have been reduced by only including individuals with 180 days (6 month) cumulative exposure before any cancer diagnosis. In addition, we excluded all individuals who had gastric cancer within a year after inclusion and also stratified the analyses by duration of use; which still showed increased risks (respectively SIR=1.61 and

SIR=2.19 among those with an exposure duration between 1 and 3 years). These increased risks may not be entirely explained by reverse causality and detection bias alone – since most cases of gastric cancer are believed to be detected within 1 year after onset of symptoms. There are no significant waiting times or socio-economic differences in access to endoscopy in Sweden, and no obvious differences were found between tumor stages (potential detection bias),(11) or anatomical locations between the PPI-exposed cohort and the background population (Appendix 3).

The fact that the risk seems to go down with a longer-duration of use is probably because PPI is beneficial for most individuals with known risk factors for cancer – yet further research seems needed for those on maintenance therapy without gastrointestinal indications.

Although our follow-up should ideally have been longer, our maximal follow-up time is 7.5 years, which is remarkably longer than all studies included in the 2 meta-analyses assessing pre-malignant lesions based on randomized controlled trials (6-36 months as maximal follow-up).(2, 6) Only 2 smaller cohort studies had longer follow-up,(1, 10) and these were included in the only meta-analysis evaluating gastric cancer risk based on observational studies.(5) We also limited our study to maintenance use, defined as at least 180 days of exposure, which is a commonly used approach.(6) Compliance and over-the-counter availability of some PPIs (Esomeprazole, Lansoprazole and Pantoprazole – only in pharmacy, not in retail sale) may have resulted in different actual dosages. H2RA were not available over-the-counter during the study period. Therefore, and because of lack of exposure data before the

study period, we cannot be certain that the registered dosage reflects the actual dosage.

The increased risks of gastric cancer among individuals with *Helicobacter pylori* infection and peptic ulcers were anticipated, since both these conditions are risk factors for gastric cancer, but the associations were stronger than expected, suggesting an additional etiological role of the PPI-use. Importantly, there was evidence of a substantially increased risk in individuals exposed to conditions not known to increase the risk of gastric cancer, including gastro-esophageal reflux. Moreover, increased risks were found even among participants without any gastrointestinal indications who were exposed only to maintenance treatment with aspirin or other NSAIDs, although these drugs are expected to decrease the risk of gastric cancer.(12, 13) The higher risk of gastric cancer in the younger age group may be related to a recently described increase in atrophic gastritis in Sweden, as a potential consequence of the stabilizing sero-prevalence of *Helicobacter pylori* and increased prevalence of overweight and obesity.(14)

The difference in the association with gastric cancer following maintenance therapy with PPIs and H2RAs could be explained simply by the fact that PPIs are more potent than H2RAs in inhibiting acid secretion.(15) PPIs block the gastric proton pumps, while H2RAs compete with histamine for the histamine-2 receptors. At recommended dosages, PPIs induce a more profound and prolonged acid inhabitation than H2RAs. Moreover, the acid inhibition of PPIs also increases over time, while the effects of H2RAs fade.(15)

There are several mechanisms that might explain an association between PPI-use and gastric cancer. Histopathological changes occur due to the blockage of the normal gastric acid secretion, leading to over-stimulation (hypergastrinemia) that might cause hyperproliferation of the gastric mucosa, chronic hypochlorhydria (reduction of hydrochloric acid in the gastric juice), chronic inflammation and disappearance of normal mucosal glands and their replacement by intestinal glands, and possible gastric atrophy.(6) All these mucosal changes could promote gastric carcinogenesis. The blocked gastric acid secretion could decrease physiological defense mechanisms against pathogenic bacteria.(7, 8) Diarrhea is for example a well-known side-effect of PPI-use, which is often due to infection with the bacteria *Clostridium difficile*, non-thyphoid *Salmonella species* or *Campylobacter jejuni.(7)* This reduced bacterial defense mechanism might, in the long term, result in chronic inflammation and eventually gastric cancer development. Decreased gastric acidity may also result in increased bacterial colonization, including non-gastric microorganisms, and a greater number of bacteria that produce nitrosamines, which are well-established carcinogens of the gastric mucosa.(5, 16)

The results of a single study, even of this size, cannot determine causality. Yet, the effect size, consistency across indications (especially those not associated with an increased risk of gastric cancer), lack of association with H2RAs, and plausible carcinogenic mechanisms mean that maintenance PPI-use cannot be dismissed as a potentially independent risk factor for gastric

cancer. A higher awareness for gastric cancer could potentially be considered in maintenance users of PPI, especially if known underlying risk factors are present. Thus, these findings need confirmation in future investigations, especially considering the generalizability of the results to populations with higher incidence of gastric cancer or other distribution of risk factors.

To conclude, this large and population-based cohort study provides evidence of a substantially increased risk of gastric cancer following maintenance use of PPIs *per se*. The consistency across sexes, age groups, and indication groups, including indications that do not increase the risk of gastric cancer, support the overall finding. If confirmed in further research, especially among those without gastrointestinal risk factors, these findings challenge a broad use of long-term PPI-use, particularly for conditions where the indication is weak.

Methods

Design

This was a nationwide Swedish population-based cohort study designed to examine the risk of gastric cancer in individuals exposed to maintenance therapy with PPIs (and to maintenance use of H2RAs), compared to the Swedish background population of the same sex, age and calendar period (7.1-7.6 million adults).(17)

Only adults (at least 18 years) without a history of any cancer were included. The participants were followed up from the first prescription of a PPI (or H2RA) during the period 1st July 2005 to 31st December 2012. The data were derived from high-quality and nationwide Swedish registries and information on individuals was linked between the registries by means of the unique Swedish personal identity number.(18) The study cohort included all Swedish residents who received at least one dispensed prescription of commonly prescribed drugs (listed in Appendix 1) between 1st July 2005 and 31st December 2014 (with follow-up for cancer until 31st December 2012). The study was approved by the Regional Ethical Review Board in Stockholm, and informed consent was not required (2014/1291-31/4).

Patient involvement

The Swedish patient organization for cancer of the esophagus, stomach, liver, and pancreas was involved in supporting the present study (www.palema.org). The development of the research question and outcome measures were informed by patients' priorities, experience, and preferences. The results will be disseminated to study participants by means of patient

organizations. Patients are thanked in the acknowledgements.

Exposure

The study exposure was maintenance therapy with a PPI (or an H2RA) according to the Swedish Prescribed Drug Registry, defined as a cumulative defined daily dose (DDD) of at least 6 months (≥180 days) during the study period (before a potential cancer diagnosis). The DDD was the average maintenance dose per day for a drug used for its main indication in adults, which follows the World Health Organization (WHO) definition. This cumulative DDD was estimated by adding the DDD per package, which takes both the potency and the quantity of the drug into account, and is therefore a proxy for the duration of the exposure. The Anatomical Therapeutic Chemical classification system (ATC) was used to identify codes representing PPIs (code A02BC) and H2RAs (code A02BA). The participants were divided into 3 mutually exclusive exposure groups: (1) PPI-users (≥180 days on PPIs and <180 days on H2RAs); (2) H2RA users (≥180 days on H2RAs and <180 days on PPIs); and (3) PPI and H2RA users (≥180 days on PPIs and ≥180 days on H2RAs). These medications were also available over-the-counter in Sweden, but only in small packages and at a much higher price per dose.(19) Thus, maintenance users had their medication prescribed and were therefore included in this study.

Outcome

The main outcome was a first episode of any gastric cancer according to the Swedish Cancer Registry. Gastric cancer was defined by the diagnosis codes

C16.0 (cardia) and C16.1-C16.9 (non-cardia) in the 10th version of the International Classification of Diseases (ICD), and adenocarcinoma (dominating histological type) was defined by the histology code 096.

Confounders

Age, sex, and calendar period were adjusted for in the design. Confounding by indication was evaluated by analyzing indications for PPI-use separately (see Appendix 2 for ICD and ATC codes): gastro-esophageal reflux disease, Barrett's esophagus, peptic ulcer disease, Zollinger-Ellison syndrome, gastro-duodenitis, dyspepsia/disruption of gastric function, *Helicobacter pylori* infection, *Helicobacter pylori* eradication (combined with *Helicobacter pylori* infection in the analyses), and maintenance use (≥180 days) of aspirin or other NSAIDs.

Data sources

Four nationwide Swedish registries, all maintained by the governmental National Board of Health and Welfare, provided data for the study.

The Swedish Prescribed Drug Registry started in 1st July 2005, and includes all drugs prescribed and dispensed in Sweden. Information from these prescriptions is transferred for registration monthly.(20) This registry is highly complete (patient identity data are missing in <0.3% of all items),(20) and was used to collect data on type of medication, dates of prescribing and dispensing, and DDD per package.

The Swedish Cancer Registry has at least a 96% complete registration of all cancers in Sweden since1958,(21) with 98% completeness for gastric

cancer.(22, 23) This registry was used to identify gastric cancer cases among the cohort members and the corresponding background population (exact annual numbers were available for the used age and sex categories), and to exclude individuals with a history of any cancer.

The Swedish Patient Registry has a full nationwide coverage for all diagnoses from 1987.(18) The registration of out-patient specialist care is nationwide since 2001. This registry was used to collect information on indications for PPI-use.

The Swedish Causes of Death Registry has 100% completeness for recording of dates of death since 1952.(24) This registry was used to collect date of death.

Statistical analyses

All analyses were conducted according to a pre-planned protocol. The risk of gastric cancer was compared between the exposed cohort and the entire Swedish background population of the same sex (male or female), age group (categorized as 18-39, 40-49, 50-59, 60-69 or ≥70 years), and calendar period (2005-2006, 2007-2009 or 2010-2012). Standardized incidence ratios (SIRs) and 95% confidence intervals (CIs) were calculated by dividing the observed number of gastric cancer cases with the expected number, accounting for changes in age and calendar categories.(25) The expected numbers were derived from the Swedish Cancer Registry and Statistics Sweden.(17) Follow-up time was calculated from the dispense date of the first prescription of PPI (or H2RA) within the study period, until death, any cancer, or end of the study period (December 31, 2012), whichever occurred first. Subgroup analyses

were stratified for sex and age. Stratified analyses were performed for each indication whenever at least 10,000 exposed individuals were identified with this indication. To assess reverse causality (protopathic bias), a sensitivity analysis was conducted excluding all cancer cases occurring within one year of the start of the study. Duration of PPI-use was estimated based on the total DDD per package prescribed before any cancer diagnosis, and was categorized into <1.0 year, 1.0-2.9 years, 3.0-4.9 years or ≥5 years.

The attributable fraction (AF) and the population attributable fraction (PAF) were calculated to estimate the proportion of gastric cancer in PPI-users and the total population, respectively (assuming causality). The following formulas were used: AF=(SIR-1)/SIR and PAF=p(SIR-1)/(p(SIR-1)+1), where p represents the prevalence of PPI maintenance use in the population (based on the population size in 2009).(17)

There were no missing data on exposures, outcomes, age, sex or calendar period. When no information was available for the indication for PPI-use (25.0%), the indication was considered absent.

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Competing interest statement: There are no competing interests.

Data sharing statement: We are willing to share data upon request after ethical approval has been approved by the relevant committee and the governmental agencies that maintain the data.

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Figure 1: Flowchart of the construction of the cohort exposed to maintenance therapy with proton pump inhibitors (PPIs) and/or H2 receptor antagonists (H2RAs).

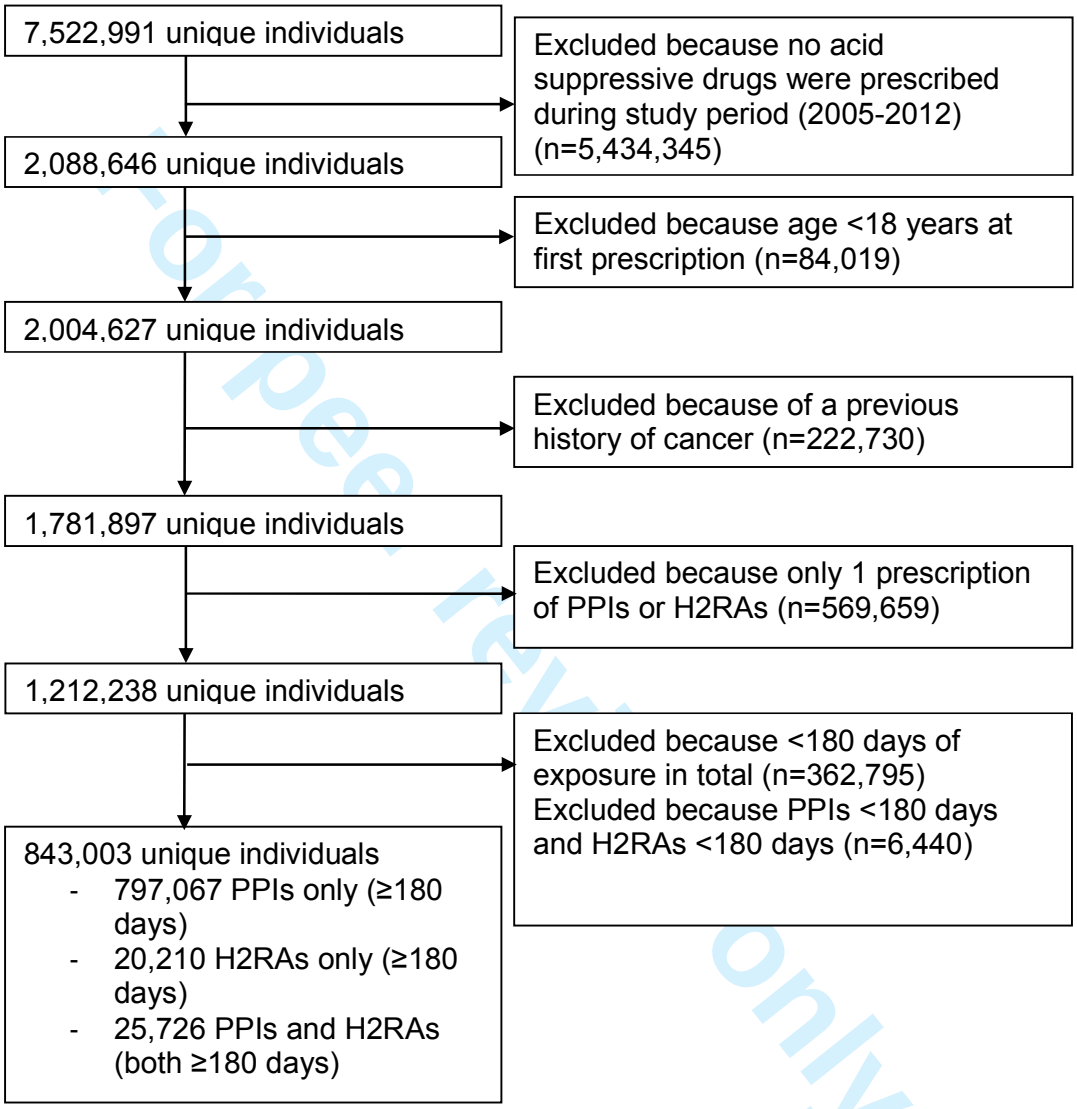

Table 1: Descriptive characteristics of the study cohort exposed to maintenance therapy with proton pump inhibitors (PPIs) and/or H2 receptor antagonists (H2RAs).

	PPIs only *Number (%)*	H2RAs only *Number (%)*	PPIs and H2RAs *Number (%)*
Total	797,067	20,210	25,726
Sex			
Men	330,885 (41.5)	8,076 (40.0)	9,778 (38.0)
Women	466,182 (58.5)	12,134 (60.0)	15,948 (62.0)
Age (years)			
<40	89,231 (11.2)	1,885 (9.3)	1,822 (7.1)
40-49	104,003 (13.1)	2,461 (12.2)	3,268 (12.7)
50-59	155,963 (19.6)	3,971 (19.7)	5,878 (22.9)
60-69	177,606 (22.3)	4,563 (22.6)	6,354 (24.7)
≥70	270,264 (33.9)	7,330 (36.3)	8,404 (32.7)
Calendar period			
2005-2006	446,068 (56.0)	16,550 (81.9)	23,066 (90.0)
2007-2009	222,257 (27.9)	2,670 (13.2)	2,304 (9.0)
2010-2012	128,742 (16.2)	990 (4.9)	356 (1.4)
Indications			
Gastro-esophageal reflux	201,868 (25.3)	2,073 (10.3)	6,727 (26.2)
Barrett's esophagus	6,044 (0.8)	17 (0.1)	172 (0.7)
Peptic ulcers	79,597 (10.0)	1,226 (6.1)	2,633 (10.2)
Zollinger-Ellison syndrome	31 (0.0)	0 (0.0)	3 (0.01)
Gastro-duodenitis	104,955 (13.2)	1,321 (6.5)	3,695 (14.4)
Dyspepsia	43,901 (5.5)	649 (3.2)	1,664 (6.5)
Helicobacter pylori	58,366 (7.3)	641 (3.2)	2,041 (7.9)
Long-term NSAID use	241,958 (30.4)	5,012 (24.8)	9,628 (37.4)
Long-term aspirin use	277,128 (34.8)	6,533 (32.3)	9,929 (38.6)
Number of indications			
0	199,608 (25.0)	7,919 (39.2)	5,402 (21.0)
1	330,027 (41.4)	8,487 (42.0)	10,320 (40.1)
≥2	267,432 (33.6)	3,804 (18.8)	10,004 (38.9)
Gastric cancer			
All	2,219 (0.28)	12 (0.06)	62 (0.24)
Adenocarcinoma	1,943 (0.24)	10 (0.05)	54 (0.21)
Cardia	567 (0.07)	1 (0.00)	13 (0.05)
Non-cardia	1,652 (0.21)	11 (0.05)	49 (0.19)
Years of follow-up			
Total	3,866,836	116,015	163,519
Mean	4.9	5.7	6.4
Cumulative defined daily dosages (DDD), estimated number of days			
Median	856	450	1741
Interquartile range	356-2102	270-990	1043-2732
Mortality			
Total	137,619 (17.3)	3,915 (19.4)	3,812 (14.8)
In gastric cancer cases	1,473 (66.4)	9 (75.0)	39 (62.9)

Table 2: Standardized incidence ratios (SIRs) by age, sex and calendar period and 95% confidence intervals (CIs) of gastric cancer in all individuals exposed to proton pump inhibitors (PPIs), stratified by sex and age-group.

	Gastric cancer		Gastric adenocarcinoma		Cardia cancer		Non-cardia gastric cancer	
	Number of cases	*SIR (95% CI)*	*Number of cases*	*SIR (95% CI)*	*Number of cases*	*SIR (95% CI)*	Number of cases	SIR (95% CI)
Total	2,219	3.38 (3.25-3.53)	1,943	3.38 (3.23-3.53)	567	3.55 (3.27-3.86)	1,652	3.33 (3.17-3.50)
Sex								
Men	1,296	3.65 (3.45-3.85)	1,166	3.66 (3.45-3.88)	437	3.84 (3.48-4.21)	859	3.56 (3.33-3.81)
Women	923	3.07 (2.87-3.28)	777	3.02 (2.82-3.25)	130	2.84 (2.38-3.38)	793	3.11 (2.99-3.34)
Age, years								
<40	36	22.76 (15.94-31.52)	27	25.30 (16.67-36.80)	7	27.33 (10.95-56.30)	29	21.88 (14.65-31.43)
40-49	94	7.36 (5.95-9.01)	76	7.76 (6.11-9.71)	24	7.54 (4.83-11.22)	70	7.30 (5.69-9.23)
50-59	303	5.63 (5.01-6.30)	263	5.82 (5.14-6.57)	94	5.50 (4.44-6.72)	209	5.69 (4.94-6.51)
60-69	576	3.85 (3.54-4.18)	501	3.81 (3.49-4.16)	191	3.94 (3.40-4.54)	385	3.81 (3.44-4.21)
≥70	1,210	2.76 (2.61-2.92)	1,076	2.77 (2.61-2.94)	251	2.77 (2.44-3.14)	959	2.76 (2.59-2.94)

Table 3: Standardized incidence ratios (SIRs) by estimated duration of use and 95% confidence intervals (CIs) of gastric cancer in all individuals exposed to proton pump inhibitors (PPIs)

Duration	Number of cases / Total	SIR (95% CI)
< 1.0 year	1,552	12.82 (12.19-13.47)
1.0-2.9 years	2,193	2.19 (1.98-2.42)
3.0-4.9 years	1,098	1.10 (0.91-1.31)
≥ 5.0 years	153	0.61 (0.52-0.72)

Table 4: Standardized incidence ratios (SIRs) by age, sex and calendar period and 95% confidence intervals (CIs) of gastric cancer in all individuals exposed to proton pump inhibitors (PPIs), stratified by indication.

	Gastric cancer		Cardia cancer		Non-cardia gastric cancer	
	Number of cases / total	SIR (95% CI)	Number of cases / total	SIR (95% CI)	Number of cases / total	SIR (95% CI)
All	2,219/ 797,067	3.38 (3.25-3.53)	567/ 797,067	3.55 (3.27-3.86)	1652/ 797,067	3.33 (3.17-3.50)
Gastro-esophageal reflux	557/ 201,868	3.04 (2.80-3.31)	175/ 201,868	3.84 (3.29-4.45)	382/ 201,868	2.78 (2.51-3.07)
Peptic ulcers	721/ 79,597	8.75 (8.12-9.41)	71/ 79,597	3.51 (2.74-4.43)	650/ 79,597	10.45 (9.66-11.28)
Gastro-duodenitis	350/ 104,955	3.68 (3.31-4.09)	63/ 104,955	2.71 (2.09-3.47)	287/ 104,955	3.99 (3.55-4.48)
Dyspepsia	136/ 43,901	3.07 (2.58-3.63)	33/ 43,901	3.10 (2.13-4.35)	103/ 43,901	3.07 (2.50-3.72)
Helicobacter pylori	440/ 58,366	9.76 (8.87-10.71)	48/ 58,366	4.19 (3.09-5.55)	392/ 58,366	11.67 (10.53-12.87)
No *Helicobacter pylori*	1,779/ 781,955	2.91 (2.78-3.05)	519/ 781,955	3.50 (3.21-3.82)	1,302/ 781,955	2.73 (2.58-2.88)
NSAIDS exposure	347/ 256,598	1.82 (1.64-2.03)	94/ 256,598	2.07 (1.67-2.53)	253/ 256,598	1.75 (1.54-1.97)
Only NSAIDS exposure	93/ 118,987	1.41 (1.14-1.73)	36/ 118,987	2.29 (1.60-3.17)	57/ 118,987	1.14 (0.86-1.48)
Aspirin exposure	872/ 293,590	2.42 (2.26-2.60)	211/ 293,590	2.73 (2.37-3.13)	588/ 293,590	2.33 (2.14-2.53)
Only aspirin exposure	252/ 126,883	1.93 (1.70-2.18)	85/ 126,883	2.73 (2.18-3.37)	169/ 126,883	1.68 (1.43-1.95)

** NSAIDs, non-steroidal anti-inflammatory drugs. One individual could have different indications, except for the group exposed only to NSAIDs or aspirin.*

Appendix 1: Description of original cohort, the "Chemoprevention of Cancer" cohort.

This cohort included all individuals residing in Sweden who received at least one dispensed prescription of one of the following commonly prescribed drugs between July 2005 and December 2014 (with corresponding ATC codes): sex hormones (G03), drugs for peptic ulcers and gastro-esophageal reflux disease (A02B), acetylsalicylic acid (B01AC06, N02BA), non-steroidal anti-inflammatory drugs (M01A), HMG CoA reductase inhibitors (C10AA, C10BA), drugs affecting bone structure and mineralization, (M05B), and antibiotics (J01AA, J01CA04, J01FA, J01MA, J01XD, J01XE, J04AB04). This cohort included approximately 85% of all Swedish residents, with especially high coverage of adults. All adult individuals receiving at least one dispensed prescription of a gastric acid inhibitor (proton pump inhibitor or H2 receptor antagonist) were included.

Appendix 2: International Classification of Diseases (ICD) and Anatomical Therapeutic Chemical classification system (ATC) codes to describe the indication for gastric acid inhibitors.

	ICD-7 *(Since 1955)*	**ICD-8** *(Since 1968)*	**ICD-9** *(Since 1987)*	**ICD-10** *(Since 1997)*
Reflux codes	784,3; 539,11-539,12; 560,4	530,93-530,94; 551,3; 784,3	530B-C; 553D; 787B	K20-21; K44; R12
Barrett's esophagus	-	-	-	K227
Peptic ulcer disease	540-541	531-533	531-533	K25-K27
Zollinger-Ellison syndrome / Hypergastrinemia	-	-	251F	E16.4
Gastro-duodenitis	543	535	535	K29
Dyspepsia/disruption of gastric function	544	536	536	K30-K31
Helicobacter pylori infection	-	-	-	B96.8 or B98.0
	ATC			
Non-steroidal anti-inflammatory drugs	M01A			
Aspirin	B01AC06, N02BA			
Helicobacter pylori eradication	A02BD, or A02BC (PPI) in combination with 2 out of 3 antibiotics (Amoxicillin [J01CA04]; Clarithromycin [J01FA09], or metronidazole [J01XD]); or combined with metronidazole [J01XD], doxycycline [J01AA02] and bismuth [A02BX05]; or with amoxicillin [J01CA04] and levofloxacin [J01MA12].			

Note: an indication was considered present if the ICD code occurred at least once in the in- or out-Patient Registry. NSAIDs and Aspirin were only considered if the cumulative dosage was ≥180 days.

Appendix 3: Tumor stage and anatomical locations of gastric cancer in the cohort exposed to maintenance use of proton pump inhibitors (PPIs) and in the Swedish background population of the same age.

	PPI maintenance cohort	Background population
	Number (%)	Number (%)
Total	2219 (100.0)	5821 (100.0)
Tumor stage - Cardia		
0-1	72 (12.7)	134 (8.9)
2	36 (6.4)	80 (5.3)
3	207 (36.5)	480 (31.9)
4	145 (25.6)	496 (33.0)
unclear/incomplete	107 (18.9)	315 (20.9)
Tumor stage - Non-cardia		
0-1	291 (17.6)	617 (14.3)
2	68 (4.1)	168 (3.9)
3	408 (24.7)	979 (22.7)
4	422 (25.5)	1260 (29.2)
unclear/incomplete	463 (28.0)	1292 (29.9)
Anatomical location		
Cardia	567 (25.6)	1505 (25.9)
Fundus	97 (4.4)	268 (4.6)
Corpus	389 (17.5)	1040 (17.9)
Antrum	398 (17.9)	915 (15.7)
Pylorus	157 (7.1)	303 (5.2)
Minor curvature	74 (3.3)	173 (3.0)
Major curvature	33 (1.5)	93 (1.6)
Overgrowth of unknown origin	2 (0.1)	15 (0.3)
Unspecified	502 (22.6)	1509 (25.9)

STROBE Statement—Checklist of items that should be included in reports of ***cohort studies***

	Item No	Recommendation	Page
Title and abstract	1	(*a*) Indicate the study's design with a commonly used term in the title or the abstract	p 1-2 (abstract & title)
		(*b*) Provide in the abstract an informative and balanced summary of what was done and what was found	p 2
Introduction			
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported	p 4-5
Objectives	3	State specific objectives, including any prespecified hypotheses	p 4-5
Methods			
Study design	4	Present key elements of study design early in the paper	p 6-10
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	p 6-10
Participants	6	(*a*) Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up	p 6-10
		(*b*) For matched studies, give matching criteria and number of exposed and unexposed	n/a
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable	p 6-10 + appendix
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	p 6-10
Bias	9	Describe any efforts to address potential sources of bias	p 6-10
Study size	10	Explain how the study size was arrived at	Fig 1
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why	p 6-10
Statistical methods	12	(*a*) Describe all statistical methods, including those used to control for confounding	p 9-10
		(*b*) Describe any methods used to examine subgroups and interactions	p 9-10
		(*c*) Explain how missing data were addressed	p 9-10
		(*d*) If applicable, explain how loss to follow-up was addressed	n/a
		(*e*) Describe any sensitivity analyses	p 9-10
Results			
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed	Fig 1
		(b) Give reasons for non-participation at each stage	Fig 1
		(c) Consider use of a flow diagram	Fig 1
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders	p 11, Table 1
		(b) Indicate number of participants with missing data for each variable of interest	n/a
		(c) Summarise follow-up time (eg, average and total amount)	Table 1
Outcome data	15*	Report numbers of outcome events or summary measures over time	Tables 1-3
Main results	16	(*a*) Give unadjusted estimates and, if applicable, confounder-adjusted	Tables 2-3

		estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included	
		(*b*) Report category boundaries when continuous variables were categorized	Table 1-2
		(*c*) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period	Discussion p 13
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses	p 11-14
Discussion			
Key results	18	Summarise key results with reference to study objectives	p 15-19
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias	p 15-19
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	p 15-19
Generalisability	21	Discuss the generalisability (external validity) of the study results	p 15-19
Other information			
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	p 3, p 10

*Give information separately for exposed and unexposed groups.

Note: An Explanation and Elaboration article discusses each checklist item and gives methodological background and published examples of transparent reporting. The STROBE checklist is best used in conjunction with this article (freely available on the Web sites of PLoS Medicine at http://www.plosmedicine.org/, Annals of Internal Medicine at http://www.annals.org/, and Epidemiology at http://www.epidem.com/). Information on the STROBE Initiative is available at http://www.strobe-statement.org.

BMJ Open

Maintenance therapy with proton pump inhibitors and risk of gastric cancer: a nationwide population-based cohort study in Sweden

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Title: Maintenance therapy with proton pump inhibitors and risk of gastric cancer: a nationwide population-based cohort study in Sweden

Running title: Proton pump inhibitors and risk of gastric cancer

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Abstract (299 words)

Objective: Proton pump inhibitors (PPIs) are among the most commonly prescribed drugs. Concerns have been raised about a potentially increased risk of gastric cancer following long-term use. Our aim is to assess the risk of gastric cancer associated with PPI use, taking into account underlying indications.

Design: Population-based cohort study. Standardized incidence ratios (SIRs) and 95% confidence intervals (CIs) were calculated to compare the risk of gastric cancer among long-term PPI-users with the corresponding background population, while taking confounding by indication into account.

Setting: Population-based study in Sweden (2005-2012)

Participants: this study included virtually all adults residing in Sweden exposed to maintenance therapy with PPIs.

Exposure/Intervention: maintenance use of PPIs, defined as at least 180 days during the study period. Maintenance use of H2-receptor antagonist was evaluated for comparison reasons.

Outcome measures: gastric cancer (cardia and non-cardia), and subgroup analysis for gastric adenocarcinoma, as defined by the Swedish Cancer Register.

Results: Among 797,067 individuals on maintenance PPI therapy, the SIR of gastric cancer was over 3-fold increased (SIR=3.38, 95% CI 3.23-3.53). Increased SIRs were found in both sexes and all age groups, but were especially increased among PPI-users younger than 40 years (SIR=22.76, 95% CI 15.94-31.52). Increased SIRs were found for each indication studied,

including those without an association with gastric cancer, e.g. gastro-esophageal reflux (SIR=3.04, 95% CI 2.80-3.31), and those with a supposedly decreased risk, e.g. aspirin users (SIR=1.93, 95% CI 1.70-2.18). The association was similar for cardia and non-cardia gastric cancer. Analyses restricted to adenocarcinoma showed similar results to those for all gastric cancers. Long-term users of histamine-2 receptor antagonists, which have the same indications as PPIs, were not at any increased risk.

Conclusions: Long-term PPI-use might be an independent risk factor for gastric cancer. This challenges broad maintenance PPI therapy, particularly if the indication is weak.

Article summary Strengths and limitations of this study

- Population-based and nationwide design based on contemporary use of proton-pump inhibitors (PPIs) – resulting in sufficient power to assess underlying indications (to assess confounding by indication).
- To our knowledge, this is the largest study to date assessing the association between PPIs and gastric cancer
- The findings are standardized for age, sex – which are often described the major confounding factors in epidemiologic studies - and calendar time. Yet, other confounders could not be taken into account because the information was not available for the background population.
- Exposure information is based on the Swedish Prescribed Drug Registry, which is initiated in July 2005 and has a complete nationwide coverage.
- Although this study provides some evidence for an association between PPI use and gastric cancer beyond gastro-intestinal indications of use, conclusions considering causality can never be drawn based on a single study.

Introduction

Proton pump inhibitors (PPIs), introduced in the 1980s, are potent gastric acid suppressors that reduce gastric acidity.(1-3) PPIs are among the most prescribed drugs globally,(4) used for healing peptic ulcers, counteracting gastro-esophageal reflux, eradicating *Helicobacter pylori* (in combination with antibiotics) and preventing primary or recurrent peptic ulcers, e.g. in individuals exposed to aspirin or other non-steroidal anti-inflammatory drugs (NSAIDs) or with Zollinger-Ellison syndrome (a gastrin-secreting pancreatic tumor). However, it has been suggested that long-term PPI-use increases the risk of pre-malignant gastric lesions (e.g. polyps, atrophy, and metaplasia) and gastric cancer.(2, 5, 6) Gastric acid secretion blockage may disrupt the gastric microbiome, interfere with nitrosamine formation, cause chronic atrophic gastritis and increase gastrin serum levels, which can all contribute to gastric cancer development.(2, 5, 7, 8). The effect of PPI use on the gut microbiome may even be more prominent than the effects of antibiotics.(9) Among three recent meta-analyses on the topic, one found no association between long-term PPI-use and pre-malignant gastric lesions, based on six randomized controlled trials (1789 patients in total).(2) The second included an additional trial (2343 patients in total), and found no evidence of gastric tumor development in PPI-users with atrophy or intestinal metaplasia, while an increased risk of gastric hyperplasia was indicated.(6) The third, based on 11 observational studies (94,558 participants), reported a 40% increase of gastric cancer among PPI-users.(5) However, the impact of confounding by indication remains unknown. The present study aimed to assess the risk of gastric cancer in long-term PPI-users in a population-based design, while

taking confounding by indication for such treatment into account. For comparison reasons, use of histamine 2 receptor antagonists (H2RAs), which are used for similar indications as PPIs, was also studied.

Methods

Design

This was a nationwide Swedish population-based cohort study designed to examine the risk of gastric cancer in individuals exposed to maintenance therapy with PPIs (and to maintenance use of H2RAs), compared to the Swedish background population of the same sex, age and calendar period (7.1-7.6 million adults).(10)

Only adults (at least 18 years) without a history of any cancer were included. The participants were followed up from the first prescription of a PPI (or H2RA) during the period 1st July 2005 to 31st December 2012. The data were derived from high-quality and nationwide Swedish registries and information on individuals was linked between the registries by means of the unique Swedish personal identity number.(11) The source cohort included all Swedish residents who received at least one dispensed prescription of commonly prescribed drugs (listed in Appendix 1) between 1st July 2005 and 31st December 2014 (with follow-up for cancer until 31st December 2012). The study was approved by the Regional Ethical Review Board in Stockholm, and informed consent was not required (2014/1291-31/4).

Patient involvement

The Swedish patient organization for cancer of the esophagus, stomach, liver, and pancreas was involved in supporting the present study (www.palema.org). The development of the research question and outcome measures were informed by patients' priorities, experience, and preferences. The results will be disseminated to study participants by means of patient

organizations. Patients are thanked in the acknowledgements.

Exposure

The study exposure was maintenance therapy with a PPI (or an H2RA) according to the Swedish Prescribed Drug Registry, defined as a cumulative defined daily dose (DDD) of at least 6 months (≥180 days) during the study period (before a potential cancer diagnosis). The DDD was the average maintenance dose per day for a drug used for its main indication in adults, which follows the World Health Organization (WHO) definition. This cumulative DDD was estimated by adding the DDD per package, which takes both the potency and the quantity of the drug into account, and is therefore a proxy for the duration of the exposure. The Anatomical Therapeutic Chemical classification system (ATC) was used to identify codes representing PPIs (code A02BC) and H2RAs (code A02BA). The participants were divided into 3 mutually exclusive exposure groups: (1) PPI-users (≥180 days on PPIs and <180 days on H2RAs); (2) H2RA users (≥180 days on H2RAs and <180 days on PPIs); and (3) PPI and H2RA users (≥180 days on PPIs and ≥180 days on H2RAs). These medications were also available over-the-counter in Sweden, but only in small packages and at a much higher price per dose.(12) Thus, maintenance users had their medication prescribed and were therefore included in this study.

Outcome

The main outcome was a first episode of any gastric cancer according to the Swedish Cancer Registry. Gastric cancer was defined by the diagnosis codes

C16.0 (cardia) and C16.1-C16.9 (non-cardia) in the 10th version of the International Classification of Diseases (ICD), and adenocarcinoma (dominating histological type) was defined by the histology code 096.

Confounders

Age, sex, and calendar period were adjusted for in the design. Confounding by indication was evaluated by analyzing indications for PPI-use separately (see Appendix 2 for ICD and ATC codes): gastro-esophageal reflux disease, Barrett's esophagus, peptic ulcer disease, Zollinger-Ellison syndrome, gastro-duodenitis, dyspepsia/disruption of gastric function, *Helicobacter pylori* infection, *Helicobacter pylori* eradication (combined with *Helicobacter pylori* infection in the analyses), and maintenance use (≥180 days) of aspirin or other NSAIDs.

Data sources

Four nationwide Swedish registries, all maintained by the governmental National Board of Health and Welfare, provided data for the study.

The Swedish Prescribed Drug Registry started in 1st July 2005, and includes all drugs prescribed and dispensed in Sweden. Information from these prescriptions is transferred for registration monthly.(13) This registry is highly complete (patient identity data are missing in <0.3% of all items),(13) and was used to collect data on type of medication, dates of prescribing and dispensing, and DDD per package.

The Swedish Cancer Registry has at least a 96% complete registration of all cancers in Sweden since1958,(14) with 98% completeness for gastric

cancer.(15, 16) This registry was used to identify gastric cancer cases among the cohort members and the corresponding background population (exact annual numbers were available for the used age and sex categories), and to exclude individuals with a history of any cancer.

The Swedish Patient Registry has a full nationwide coverage for all diagnoses from 1987.(11) The registration of out-patient specialist care is nationwide since 2001. This registry was used to collect information on indications for PPI-use.

The Swedish Causes of Death Registry has 100% completeness for recording of dates of death since 1952.(17) This registry was used to collect date of death.

Statistical analyses

All analyses were conducted according to a pre-planned protocol. The risk of gastric cancer was compared between the exposed cohort and the entire Swedish background population of the same sex (male or female), age group (categorized as 18-39, 40-49, 50-59, 60-69 or ≥70 years), and calendar period (2005-2006, 2007-2009 or 2010-2012). Standardized incidence ratios (SIRs) and 95% confidence intervals (CIs) were calculated by dividing the observed number of gastric cancer cases with the expected number, accounting for changes in age and calendar categories.(18) The expected numbers were derived from the Swedish Cancer Registry and Statistics Sweden.(10) Follow-up time was calculated from the dispense date of the first prescription of PPI (or H2RA) within the study period, until death, any cancer, or end of the study period (December 31, 2012), whichever occurred first. Subgroup analyses

were stratified for sex and age. Stratified analyses were performed for each indication whenever at least 10,000 exposed individuals were identified with this indication. To assess reverse causality (protopathic bias), a sensitivity analysis was conducted excluding all cancer cases occurring within one year of the start of the study. Duration of PPI-use was estimated based on the total DDD per package prescribed before any cancer diagnosis, and was categorized into <1.0 year, 1.0-2.9 years, 3.0-4.9 years or ≥5 years.

The attributable fraction (AF) and the population attributable fraction (PAF) were calculated to estimate the proportion of gastric cancer in PPI-users and the total population, respectively (assuming causality). The following formulas were used: AF=(SIR-1)/SIR and PAF=p(SIR-1)/(p(SIR-1)+1), where p represents the prevalence of PPI maintenance use in the population (based on the population size in 2009).(10) The Chi-square test was used to calculate p-values.

There were no missing data on exposures, outcomes, age, sex or calendar period. When no information was available for the indication for PPI-use (25.0%), the indication was considered absent.

Results

Study participants

In total, 797,067 individuals on maintenance PPI therapy were included in the exposed cohort, resulting in 3,866,836 person-years of follow-up (mean 4.9 years). An additional 20,210 individuals had maintenance H2RA therapy, and 25,726 had both PPI and H2RA maintenance therapy. Figure 1 presents how the study participants were selected, and Table 1 shows their characteristics. Women constituted 58.5% of the PPI cohort, and a majority (66.1%) was younger than 70 years. Maintenance therapy with aspirin (34.8%) and NSAIDs (30.4%) were the most common indications for maintenance PPI use, followed by gastro-esophageal reflux (25.3%), gastro-duodenitis (13.2%) and peptic ulcer (10.0%). *Helicobacter pylori* was the indication in 7.3% of all participants, and dyspepsia in 5.5%. Barrett's esophagus and Zollinger-Ellison syndrome were rare indications (<1%) and therefore not eligible for separate analysis. A third (33.4%) of the participants had more than one of the listed indications, and was therefore included in more than one indication group. Overall mortality was slightly lower among PPI users (17.3%) compared to H2RA maintenance users (19.4%)(p<0.001), while mortality in gastric cancer patients did not differ significantly (p=0.782).

Proton pump inhibitor use and overall risk of gastric cancer

Among all participants exposed to maintenance PPIs, 2,219 (0.28%) developed gastric cancer during follow-up. Of these, 1,652 (74.4%) had non-cardia gastric cancer and 1,943 (87.6%) had adenocarcinoma (Table 1). Long-term PPI-users were at more than a 3-fold increased SIR of gastric

cancer of any type (SIR=3.38, 95% CI 3.25-3.53) and gastric adenocarcinoma (SIR=3.38, 95% CI 3.23-3.53) (Table 2). After excluding early gastric cancers, the risk remained increased (SIR=1.61, 95% CI 1.51-1.71). The risk of gastric cancer was highest among individuals receiving PPI for shorter than 1 year (SIR=12.82, 95% CI 12.19-13.47), but there was also evidence for an increased risk up to 3 years of PPI use, yet reduced risks for use over 5 years (Table 3). The strength of association was similar for cardia (SIR=3.55, 95% CI 3.27-3.86) and non-cardia gastric cancer (SIR=3.33, 95% CI 3.17-3.50). The risk of gastric cancer was similar in men (SIR=3.65, 95% CI 3.45-3.85) and women (SIR=3.07, 95% CI 2.87-3.28). The SIR was higher in younger ages; PPI-users younger than 40 years had SIR of 22.76 (95% CI 15.94-31.52) while those who were at least 70 years old had SIR of 2.76 (95% CI 2.61-2.92) (Table 2). The subgroup analyses for sex and age showed similar results as the overall findings in separate analyses for gastric adenocarcinoma, as well as for cardia and non-cardia gastric cancer (Table 2).

Proton pump inhibitor use and risk of gastric cancer stratified by indication

The SIR of gastric cancer was increased in each of the 10 studied groups of indications for PPI therapy (Table 4). The highest SIRs were found among participants exposed to indications with a known association with gastric cancer, i.e. *Helicobacter pylori* and peptic ulcer. However, the SIRs were also increased for indications without any such association. The SIR for gastric cancer was 3.04 (95% CI 2.80-3.31) for gastro-esophageal reflux. The SIR

were increased also for indications where a decreased risk of gastric cancer was expected. The SIR was 1.93 (95% CI 1.70-2.18) among those using PPIs because of aspirin use without any other indication. The SIRs were generally similar for non-cardia gastric cancer. Regarding cardia cancer, higher SIRs than for gastric cancer were found for the indication gastro-esophageal reflux and among those exposed to aspirin or other NSAIDs, while the associations for *Helicobacter pylori* and peptic ulcer were less strong (Table 4).

Attributable risk

Of all 5,823 gastric cancer patients diagnosed during the study period, 38.1% occurred among maintenance PPI-users. Based on the overall SIR of 3.38, and a prevalence of PPI-use of 10.7% among adults, the attributable fraction among PPI-users (AF) was 70.4%, and 20.3% in the total population (PAF), assuming causality. Using the lowest SIR according to Table 4 (SIR=1.41), the corresponding proportions were 29.1% (AF) and 4.2% (PAF), respectively. Among individuals younger than 40, the prevalence of PPI-use was 3.3%, and 37.1% of all 97 gastric cancers occurred among PPI-users, corresponding to AF of 95.6% and PAF of 41.8%.

Use of histamine-2 receptor blockers only or proton pump inhibitors and histamine-2 receptor blockers and risk of gastric cancer

There were 12 and 62 cases of gastric cancer among maintenance H2RAs users only and both H2RAs and PPIs, respectively. The risk of gastric cancer was not increased in the H2RA only group (SIR=0.57, 95% CI 0.29-0.99), and

was moderately increased in the group exposed to both PPIs and H2RAs (SIR=2.09, 95% CI 1.61-2.69).

Discussion

This study provides some evidence of an increased risk of gastric cancer among maintenance PPI-users, including those who had PPI therapy for indications without any positive association with gastric cancer. Increased risks were found for both cardia and non-cardia gastric cancer, in both sexes and all age groups, although the risk was more pronounced in younger participants.

Strengths of the study include the nationwide and population-based design, the valid data on exposures, outcomes, and indications for PPI-use, and the large number of individuals exposed to maintenance treatment with PPIs. To our knowledge, this is the largest study to date on this topic,(19) and the cohort allowed analyses of differences in associations between underlying indications.

The main problems of this study are confounding, especially by indication, and reverse causality. Confounding by indication has been investigated by looking at the different indication groups, including groups without no increased risk of gastric cancer. Unfortunately, we could not identify any underlying indication for 25% of the PPI-users, and 39% of the H2RA-users. It is likely that clear indications were more readily recorded, while use for less rigorously diagnosed indications were more prone to be missing. This may indicate that PPI-users had more severe symptoms than H2RA-users, as also indicated by the higher proportion of PPI users presenting with multiple indications (34% vs. 19%). Yet, the lack of information on indication should not explain the associations for known indications – and those groups with no

apparent risk factors should be less likely to have severe gastrointestinal symptoms. Therefore, it was unexpected to still see such increased risks of gastric cancer in those groups. Unfortunately, we lacked information about some potential confounders, e.g. dietary factors, obesity, tobacco smoking and alcohol overconsumption since these are not collected in the nationwide Health Registries. However, the lack of association between H2RAs and gastric cancer argues against bias from confounding or selection from lifestyle factors or any other unknown factors. Since H2RAs were used for similar indications as PPIs (yet clearly became less popular), the lack of association between H2RAs and gastric cancer supports that the association between PPI-use and gastric cancer may be linked to the PPI medication *per se*. Yet, PPIs clearly became the first choice treatment for most indications for gastric acid suppression with almost 40 times more PPI maintenance users than H2RA maintenance users in Sweden. The high prevalence of PPI maintenance use among adults (10.7% in Sweden) also means that the large majority with recognized risk factors for gastric cancer will have received PPI treatment at some point. This clearly hampers assessment of PPI use as an independent risk factor and this may even be a larger problem in other study designs especially if indications of use cannot be assessed.

The problem of reverse causality, i.e. individuals taking PPIs because of symptoms arising from an undetected cancer, should have been reduced by only including individuals with at least 180 days (6 months) of cumulative exposure before any cancer diagnosis. In addition, we excluded all individuals who had gastric cancer within a year after inclusion and also stratified the analyses by duration of use; which still showed increased risks (respectively

SIR=1.61 and SIR=2.19 among those with an exposure duration between 1 and 3 years). These increased risks may not be entirely explained by reverse causality and detection bias alone – since most cases of gastric cancer are believed to be detected within 1 year after onset of symptoms. There are no significant waiting times or socio-economic differences in access to endoscopy in Sweden, and no obvious differences were found between tumor stages (potential detection bias),(20) or anatomical locations between the PPI-exposed cohort and the background population (Appendix 3).

The seemingly decreasing risk estimates with a longer-duration of use is probably because PPI is beneficial for most individuals with known risk factors for cancer (e.g. peptic ulcers, *H. pylori)* – yet further research seems needed for those on maintenance therapy without gastrointestinal indications (e.g. aspirin and NSAIDs users).

Although our follow-up should ideally have been longer, our maximal follow-up time is 7.5 years, which is remarkably longer than all studies included in the 2 meta-analyses assessing pre-malignant lesions based on randomized controlled trials (6-36 months as maximal follow-up).(2, 6) Only 2 smaller cohort studies had longer follow-up,(1, 19) and these were included in the only meta-analysis evaluating gastric cancer risk based on observational studies.(5) We also limited our study to maintenance use, defined as at least 180 days of exposure, which is a commonly used approach.(6) Compliance and over-the-counter availability of some PPIs (Esomeprazole, Lansoprazole and Pantoprazole – only in pharmacy, not in retail sale) may have resulted in different actual dosages. H2RA were not available over-the-counter during the study period. Therefore, and because of lack of exposure data before the

study period, we cannot be certain that the registered dosage reflects the actual dosage.

The increased risks of gastric cancer among individuals with *Helicobacter pylori* and peptic ulcers were anticipated, since both these conditions are risk factors for gastric cancer, but the associations were stronger than expected, suggesting an additional etiological role of the PPI-use. Importantly, there was evidence of a substantially increased risk in individuals exposed to conditions not known to increase the risk of gastric cancer, including gastro-esophageal reflux. Moreover, increased risks were found even among participants without any gastrointestinal indications who were exposed only to maintenance treatment with aspirin or other NSAIDs, although these drugs are expected to decrease the risk of gastric cancer.(21, 22)

The higher risk of gastric cancer in the younger age group might be related to a recently described increase in atrophic gastritis in Sweden, as a potential consequence of the stabilizing sero-prevalence of *Helicobacter pylori* and increased prevalence of overweight and obesity.(23) Increasing risks of gastric cancer in young populations have been described previously, in particular of the diffuse type of gastric adenocarcinoma. (24, 25) Gastric symptoms in this group are also often over-looked, leading to a widespread (uncontrolled) use of PPIs.(25) Additionally, gastric carcinogenesis may be accelerated in younger patients (who also have a higher likelihood of a family history of gastric cancer),(26) which may in turn contribute to a higher vulnerability to PPI's potentially harmful effects. A further exploration into the

presence and severity of gastrointestinal risk factors, age-specific effects and potential mechanistic pathways is warranted.

The difference in the association with gastric cancer following maintenance therapy with PPIs and H2RAs could be explained simply by the fact that PPIs are more potent than H2RAs in inhibiting acid secretion.(27) PPIs block the gastric proton pumps, while H2RAs compete with histamine for the histamine-2 receptors. At recommended dosages, PPIs induce a more profound and prolonged acid inhabitation than H2RAs. Moreover, the acid inhibition of PPIs also increases over time, while the effects of H2RAs fade.(27)

There are several mechanisms that might explain an association between PPI-use and gastric cancer. Already 30 years ago, animal studies showed that profound inhibition of gastric acid secretion in rodents induces gastric tumours, with secondary over-stimulation (hypergastrinemia) leading to enterochromaffin-like cell (ECL) hyperplasia as generally accepted mechanism of this carcinogenic effect.(28, 29) ECL differentiation has also been described in human gastric carcinomas, particularly in the signet ring subtype.(30, 31) Histopathological changes occur due to the blockage of the normal gastric acid secretion, leading to hypergastrinemia that might cause hyperproliferation of the gastric mucosa, chronic hypochlorhydria (reduction of hydrochloric acid in the gastric juice), chronic inflammation and disappearance of normal mucosal glands and their replacement by intestinal glands, and possible gastric atrophy.(6) Long-term PPI use has also been linked to the development of fundic gland polyps.(32) All these mucosal

changes could promote gastric carcinogenesis. The blocked gastric acid secretion could decrease physiological defense mechanisms against pathogenic bacteria.(7, 8) Diarrhea is for example a well-known side-effect of PPI-use, which is often due to infection with the bacteria *Clostridium difficile*, non-thyphoid *Salmonella species* or *Campylobacter jejuni*.*(7)* This reduced bacterial defense mechanism might, in the long term, result in chronic inflammation and eventually gastric cancer development. Decreased gastric acidity may also result in increased bacterial colonization, including non-gastric microorganisms, and a greater number of bacteria that produce nitrosamines, which are well-established carcinogens of the gastric mucosa.(5, 33) The impact of these proposed underlying mechanisms may also be person- and time-dependent, which could explain why PPIs are seemingly more harmful in for example the younger age groups.

The results of a single study, even of this size, cannot determine causality. Yet, the effect size, consistency across indications (especially those not associated with an increased risk of gastric cancer), lack of association with H2RAs, and plausible carcinogenic mechanisms mean that maintenance PPI-use cannot be dismissed as a potentially independent risk factor for gastric cancer. A higher awareness for gastric cancer could potentially be considered in maintenance users of PPI, especially if known underlying risk factors are present. Thus, these findings need confirmation in future investigations, especially considering the generalizability of the results to populations with higher incidence of gastric cancer or other distribution of risk factors.

To conclude, this large and population-based cohort study provides evidence of a substantially increased risk of gastric cancer following maintenance use of PPIs *per se*. The consistency across sexes, age groups, and indication groups, including indications that do not increase the risk of gastric cancer, support the overall finding. If confirmed in further research, especially among those without gastrointestinal risk factors, these findings challenge a broad use of long-term PPI-use, particularly for conditions where the indication is weak.

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Data sharing statement: We are willing to share data upon request after ethical approval has been approved by the relevant committee and the governmental agencies that maintain the data.

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Figure 1: Flowchart of the construction of the cohort exposed to maintenance therapy with proton pump inhibitors (PPIs) and/or H2 receptor antagonists (H2RAs).

Table 1: Descriptive characteristics of the study cohort exposed to maintenance therapy with proton pump inhibitors (PPIs) and/or H2 receptor antagonists (H2RAs).

	PPIs only *Number (%)*	H2RAs only *Number (%)*	PPIs and H2RAs *Number (%)*
Total	797,067	20,210	25,726
Sex			
Men	330,885 (41.5)	8,076 (40.0)	9,778 (38.0)
Women	466,182 (58.5)	12,134 (60.0)	15,948 (62.0)
Age (years)			
<40	89,231 (11.2)	1,885 (9.3)	1,822 (7.1)
40-49	104,003 (13.1)	2,461 (12.2)	3,268 (12.7)
50-59	155,963 (19.6)	3,971 (19.7)	5,878 (22.9)
60-69	177,606 (22.3)	4,563 (22.6)	6,354 (24.7)
≥70	270,264 (33.9)	7,330 (36.3)	8,404 (32.7)
Calendar period			
2005-2006	446,068 (56.0)	16,550 (81.9)	23,066 (90.0)
2007-2009	222,257 (27.9)	2,670 (13.2)	2,304 (9.0)
2010-2012	128,742 (16.2)	990 (4.9)	356 (1.4)
Indications			
Gastro-esophageal reflux	201,868 (25.3)	2,073 (10.3)	6,727 (26.2)
Barrett's esophagus	6,044 (0.8)	17 (0.1)	172 (0.7)
Peptic ulcers	79,597 (10.0)	1,226 (6.1)	2,633 (10.2)
Zollinger-Ellison syndrome	31 (0.0)	0 (0.0)	3 (0.01)
Gastro-duodenitis	104,955 (13.2)	1,321 (6.5)	3,695 (14.4)
Dyspepsia	43,901 (5.5)	649 (3.2)	1,664 (6.5)
Helicobacter pylori	58,366 (7.3)	641 (3.2)	2,041 (7.9)
Long-term NSAID use	241,958 (30.4)	5,012 (24.8)	9,628 (37.4)
Long-term aspirin use	277,128 (34.8)	6,533 (32.3)	9,929 (38.6)
Number of indications			
0	199,608 (25.0)	7,919 (39.2)	5,402 (21.0)
1	330,027 (41.4)	8,487 (42.0)	10,320 (40.1)
≥2	267,432 (33.6)	3,804 (18.8)	10,004 (38.9)
Gastric cancer			
All	2,219 (0.28)	12 (0.06)	62 (0.24)
Adenocarcinoma	1,943 (0.24)	10 (0.05)	54 (0.21)
Cardia	567 (0.07)	1 (0.00)	13 (0.05)
Non-cardia	1,652 (0.21)	11 (0.05)	49 (0.19)
Years of follow-up			
Total	3,866,836	116,015	163,519
Mean	4.9	5.7	6.4
Cumulative defined daily dosages (DDD), estimated number of days			
Median	856	450	1741
Interquartile range	356-2102	270-990	1043-2732
Mortality			
Total	137,619 (17.3)	3,915 (19.4)	3,812 (14.8)
In gastric cancer cases	1,473 (66.4)	9 (75.0)	39 (62.9)

Table 2: Standardized incidence ratios (SIRs) by age, sex and calendar period and 95% confidence intervals (CIs) of gastric cancer in all individuals exposed to proton pump inhibitors (PPIs), stratified by sex and age-group.

	Gastric cancer		Gastric adenocarcinoma		Cardia cancer		Non-cardia gastric cancer	
	Number of cases	*SIR (95% CI)*	*Number of cases*	*SIR (95% CI)*	*Number of cases*	*SIR (95% CI)*	Number of cases	SIR (95% CI)
Total	2,219	3.38 (3.25-3.53)	1,943	3.38 (3.23-3.53)	567	3.55 (3.27-3.86)	1,652	3.33 (3.17-3.50)
Sex								
Men	1,296	3.65 (3.45-3.85)	1,166	3.66 (3.45-3.88)	437	3.84 (3.48-4.21)	859	3.56 (3.33-3.81)
Women	923	3.07 (2.87-3.28)	777	3.02 (2.82-3.25)	130	2.84 (2.38-3.38)	793	3.11 (2.99-3.34)
Age, years								
<40	36	22.76 (15.94-31.52)	27	25.30 (16.67-36.80)	7	27.33 (10.95-56.30)	29	21.88 (14.65-31.43)
40-49	94	7.36 (5.95-9.01)	76	7.76 (6.11-9.71)	24	7.54 (4.83-11.22)	70	7.30 (5.69-9.23)
50-59	303	5.63 (5.01-6.30)	263	5.82 (5.14-6.57)	94	5.50 (4.44-6.72)	209	5.69 (4.94-6.51)
60-69	576	3.85 (3.54-4.18)	501	3.81 (3.49-4.16)	191	3.94 (3.40-4.54)	385	3.81 (3.44-4.21)
≥70	1,210	2.76 (2.61-2.92)	1,076	2.77 (2.61-2.94)	251	2.77 (2.44-3.14)	959	2.76 (2.59-2.94)

Table 3: Standardized incidence ratios (SIRs) by estimated duration of use and 95% confidence intervals (CIs) of gastric cancer in all individuals exposed to proton pump inhibitors (PPIs)

Duration	Number of cases / Total	SIR (95% CI)
< 1.0 year	1,552	12.82 (12.19-13.47)
1.0-2.9 years	2,193	2.19 (1.98-2.42)
3.0-4.9 years	1,098	1.10 (0.91-1.31)
≥ 5.0 years	153	0.61 (0.52-0.72)

Table 4: Standardized incidence ratios (SIRs) by age, sex and calendar period and 95% confidence intervals (CIs) of gastric cancer in all individuals exposed to proton pump inhibitors (PPIs), stratified by indication.

	Gastric cancer		Cardia cancer		Non-cardia gastric cancer	
	Number of cases / total	SIR (95% CI)	Number of cases / total	SIR (95% CI)	Number of cases / total	SIR (95% CI)
All	2,219/ 797,067	3.38 (3.25-3.53)	567/ 797,067	3.55 (3.27-3.86)	1652/ 797,067	3.33 (3.17-3.50)
Gastro-esophageal reflux	557/ 201,868	3.04 (2.80-3.31)	175/ 201,868	3.84 (3.29-4.45)	382/ 201,868	2.78 (2.51-3.07)
Peptic ulcers	721/ 79,597	8.75 (8.12-9.41)	71/ 79,597	3.51 (2.74-4.43)	650/ 79,597	10.45 (9.66-11.28)
Gastro-duodenitis	350/ 104,955	3.68 (3.31-4.09)	63/ 104,955	2.71 (2.09-3.47)	287/ 104,955	3.99 (3.55-4.48)
Dyspepsia	136/ 43,901	3.07 (2.58-3.63)	33/ 43,901	3.10 (2.13-4.35)	103/ 43,901	3.07 (2.50-3.72)
Helicobacter pylori	440/ 58,366	9.76 (8.87-10.71)	48/ 58,366	4.19 (3.09-5.55)	392/ 58,366	11.67 (10.53-12.87)
No *Helicobacter pylori*	1,779/ 781,955	2.91 (2.78-3.05)	519/ 781,955	3.50 (3.21-3.82)	1,302/ 781,955	2.73 (2.58-2.88)
NSAIDS exposure	347/ 256,598	1.82 (1.64-2.03)	94/ 256,598	2.07 (1.67-2.53)	253/ 256,598	1.75 (1.54-1.97)
Only NSAIDS exposure	93/ 118,987	1.41 (1.14-1.73)	36/ 118,987	2.29 (1.60-3.17)	57/ 118,987	1.14 (0.86-1.48)
Aspirin exposure	872/ 293,590	2.42 (2.26-2.60)	211/ 293,590	2.73 (2.37-3.13)	588/ 293,590	2.33 (2.14-2.53)
Only aspirin exposure	252/ 126,883	1.93 (1.70-2.18)	85/ 126,883	2.73 (2.18-3.37)	169/ 126,883	1.68 (1.43-1.95)

** NSAIDs, non-steroidal anti-inflammatory drugs. One individual could have different indications, except for the group exposed only to NSAIDs or aspirin.*

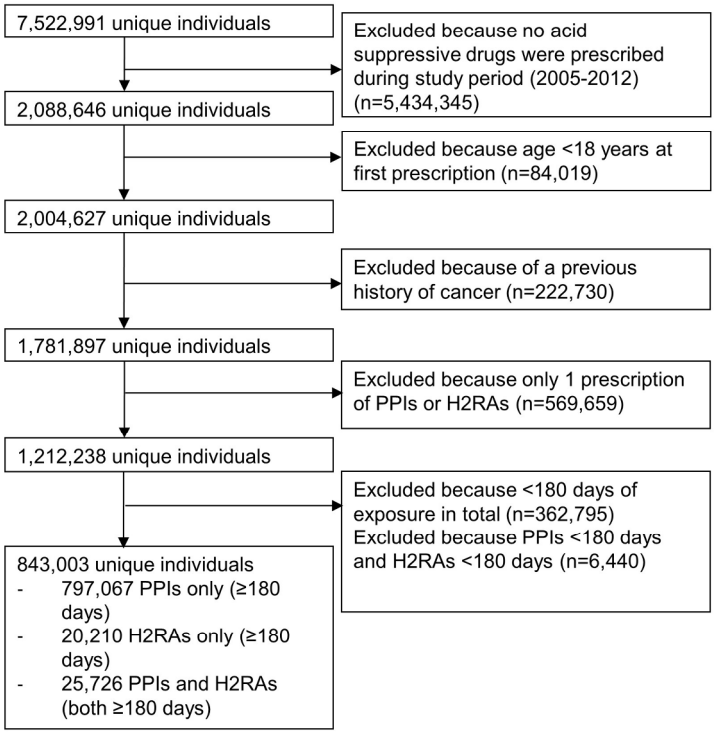

Figure 1: Flowchart of the construction of the cohort exposed to maintenance therapy with proton pump inhibitors (PPIs) and/or H2 receptor antagonists (H2RAs).

190x209mm (300 x 300 DPI)

Appendix 1: Description of original cohort, the "Chemoprevention of Cancer" cohort.

This cohort included all individuals residing in Sweden who received at least one dispensed prescription of one of the following commonly prescribed drugs between July 2005 and December 2014 (with corresponding ATC codes): sex hormones (G03), drugs for peptic ulcers and gastro-esophageal reflux disease (A02B), acetylsalicylic acid (B01AC06, N02BA), non-steroidal anti-inflammatory drugs (M01A), HMG CoA reductase inhibitors (C10AA, C10BA), drugs affecting bone structure and mineralization, (M05B), and antibiotics (J01AA, J01CA04, J01FA, J01MA, J01XD, J01XE, J04AB04). This cohort included approximately 85% of all Swedish residents, with especially high coverage of adults. All adult individuals receiving at least one dispensed prescription of a gastric acid inhibitor (proton pump inhibitor or H2 receptor antagonist) were included.

Appendix 2: International Classification of Diseases (ICD) and Anatomical Therapeutic Chemical classification system (ATC) codes to describe the indication for gastric acid inhibitors.

	ICD-7 *(Since 1955)*	**ICD-8** *(Since 1968)*	**ICD-9** *(Since 1987)*	**ICD-10** *(Since 1997)*
Reflux codes	784,3; 539,11-539,12; 560,4	530,93-530,94; 551,3; 784,3	530B-C; 553D; 787B	K20-21; K44; R12
Barrett's esophagus	-	-	-	K227
Peptic ulcer disease	540-541	531-533	531-533	K25-K27
Zollinger-Ellison syndrome / Hypergastrinemia	-	-	251F	E16.4
Gastro-duodenitis	543	535	535	K29
Dyspepsia/disruption of gastric function	544	536	536	K30-K31
Helicobacter pylori infection	-	-	-	B96.8 or B98.0
	ATC			
Non-steroidal anti-inflammatory drugs	M01A			
Aspirin	B01AC06, N02BA			
Helicobacter pylori eradication	A02BD, or A02BC (PPI) in combination with 2 out of 3 antibiotics (Amoxicillin [J01CA04]; Clarithromycin [J01FA09], or metronidazole [J01XD]); or combined with metronidazole [J01XD], doxycycline [J01AA02] and bismuth [A02BX05]; or with amoxicillin [J01CA04] and levofloxacin [J01MA12].			

Note: an indication was considered present if the ICD code occurred at least once in the in- or out-Patient Registry. NSAIDs and Aspirin were only considered if the cumulative dosage was ≥180 days.

Appendix 3: Tumor stage and anatomical locations of gastric cancer in the cohort exposed to maintenance use of proton pump inhibitors (PPIs) and in the Swedish background population of the same age.

	PPI maintenance cohort Number (%)	Background population Number (%)
Total	2219 (100.0)	5821 (100.0)
Tumor stage - Cardia		
0-1	72 (12.7)	134 (8.9)
2	36 (6.4)	80 (5.3)
3	207 (36.5)	480 (31.9)
4	145 (25.6)	496 (33.0)
unclear/incomplete	107 (18.9)	315 (20.9)
Tumor stage - Non-cardia		
0-1	291 (17.6)	617 (14.3)
2	68 (4.1)	168 (3.9)
3	408 (24.7)	979 (22.7)
4	422 (25.5)	1260 (29.2)
unclear/incomplete	463 (28.0)	1292 (29.9)
Anatomical location		
Cardia	567 (25.6)	1505 (25.9)
Fundus	97 (4.4)	268 (4.6)
Corpus	389 (17.5)	1040 (17.9)
Antrum	398 (17.9)	915 (15.7)
Pylorus	157 (7.1)	303 (5.2)
Minor curvature	74 (3.3)	173 (3.0)
Major curvature	33 (1.5)	93 (1.6)
Overgrowth of unknown origin	2 (0.1)	15 (0.3)
Unspecified	502 (22.6)	1509 (25.9)

STROBE Statement—Checklist of items that should be included in reports of ***cohort studies***

	Item No	Recommendation	Page
Title and abstract	1	(*a*) Indicate the study's design with a commonly used term in the title or the abstract	p 1-2 (abstract & title)
		(*b*) Provide in the abstract an informative and balanced summary of what was done and what was found	p 2
Introduction			
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported	p 4-5
Objectives	3	State specific objectives, including any prespecified hypotheses	p 4-5
Methods			
Study design	4	Present key elements of study design early in the paper	p 6-10
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	p 6-10
Participants	6	(*a*) Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up	p 6-10
		(*b*) For matched studies, give matching criteria and number of exposed and unexposed	n/a
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable	p 6-10 + appendix
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	p 6-10
Bias	9	Describe any efforts to address potential sources of bias	p 6-10
Study size	10	Explain how the study size was arrived at	Fig 1
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why	p 6-10
Statistical methods	12	(*a*) Describe all statistical methods, including those used to control for confounding	p 9-10
		(*b*) Describe any methods used to examine subgroups and interactions	p 9-10
		(*c*) Explain how missing data were addressed	p 9-10
		(*d*) If applicable, explain how loss to follow-up was addressed	n/a
		(*e*) Describe any sensitivity analyses	p 9-10
Results			
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed	Fig 1
		(b) Give reasons for non-participation at each stage	Fig 1
		(c) Consider use of a flow diagram	Fig 1
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders	p 11, Table 1
		(b) Indicate number of participants with missing data for each variable of interest	n/a
		(c) Summarise follow-up time (eg, average and total amount)	Table 1
Outcome data	15*	Report numbers of outcome events or summary measures over time	Tables 1-3
Main results	16	(*a*) Give unadjusted estimates and, if applicable, confounder-adjusted	Tables 2-3

		estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included	
		(*b*) Report category boundaries when continuous variables were categorized	Table 1-2
		(*c*) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period	Discussion p 13
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses	p 11-14
Discussion			
Key results	18	Summarise key results with reference to study objectives	p 15-19
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias	p 15-19
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	p 15-19
Generalisability	21	Discuss the generalisability (external validity) of the study results	p 15-19
Other information			
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	p 3, p 10

*Give information separately for exposed and unexposed groups.

Note: An Explanation and Elaboration article discusses each checklist item and gives methodological background and published examples of transparent reporting. The STROBE checklist is best used in conjunction with this article (freely available on the Web sites of PLoS Medicine at http://www.plosmedicine.org/, Annals of Internal Medicine at http://www.annals.org/, and Epidemiology at http://www.epidem.com/). Information on the STROBE Initiative is available at http://www.strobe-statement.org.